

09863803.052201

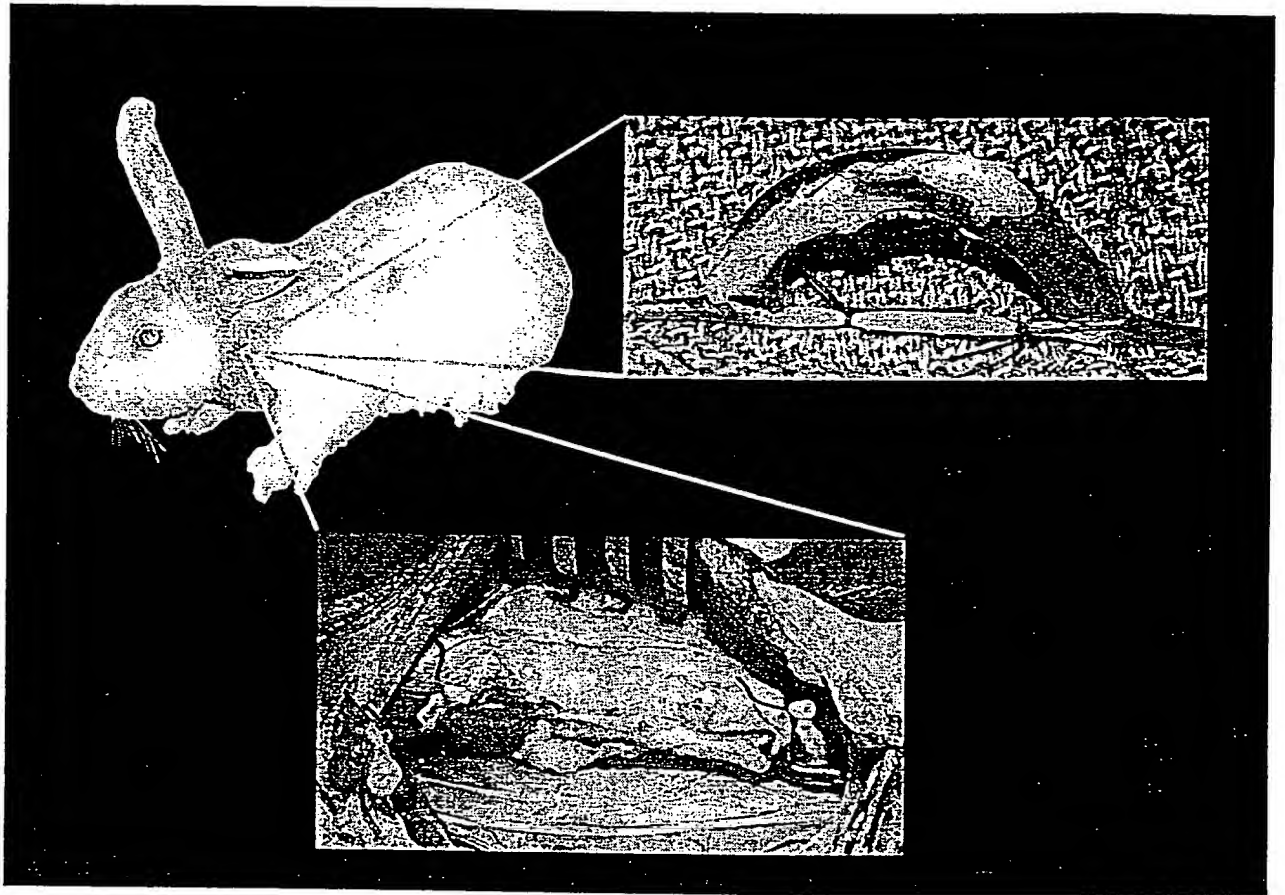


Figure 1

09863803.052201

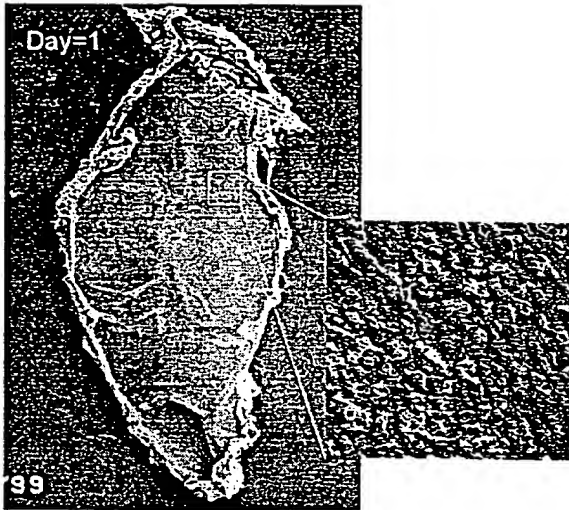


Figure 2A

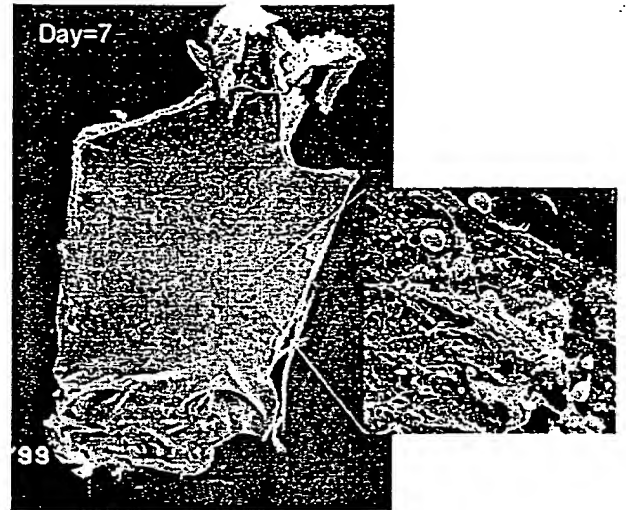


Figure 2B

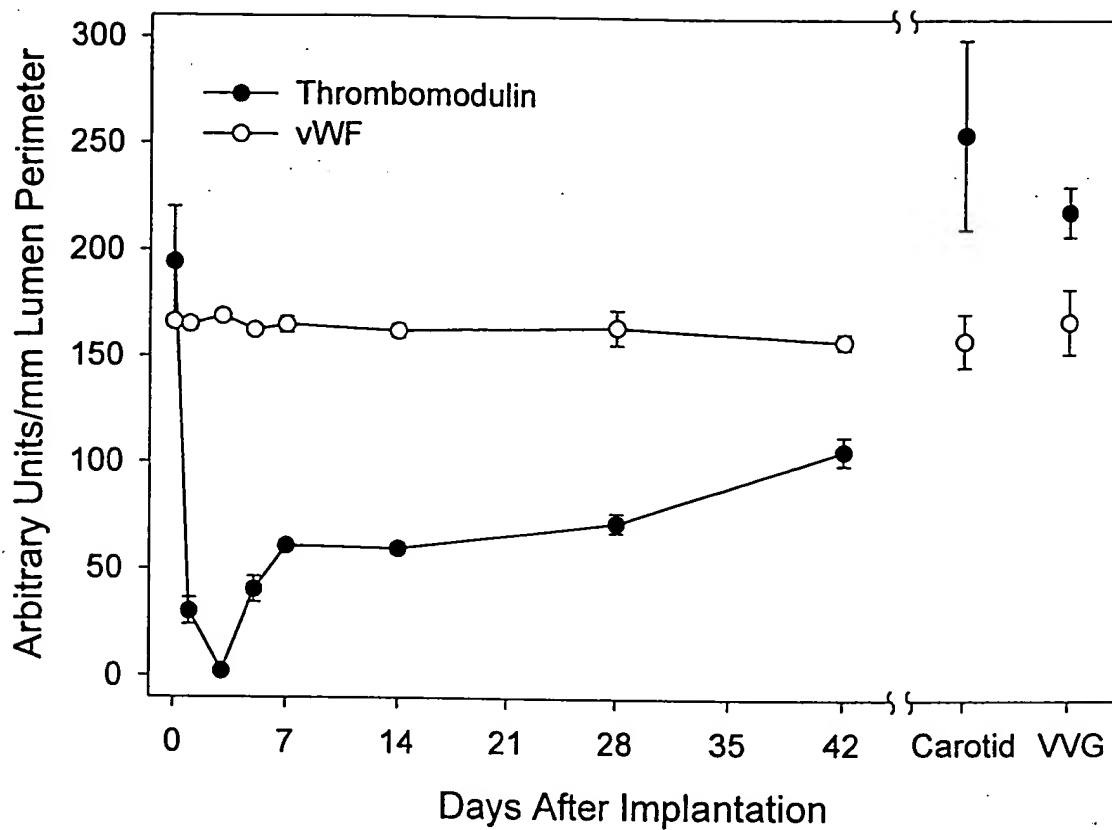


Figure 3

Fig. 4A

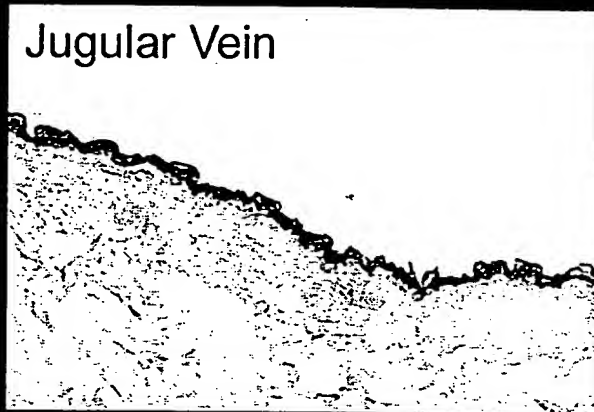
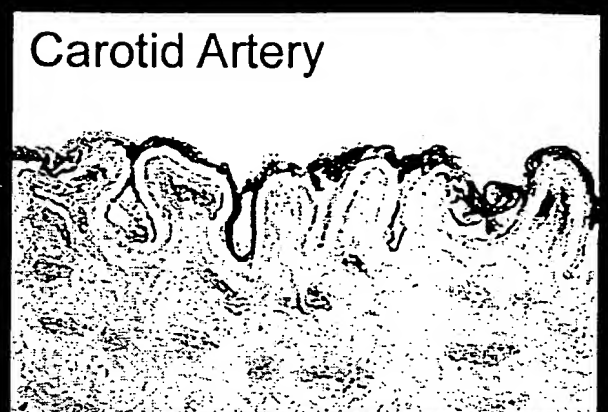
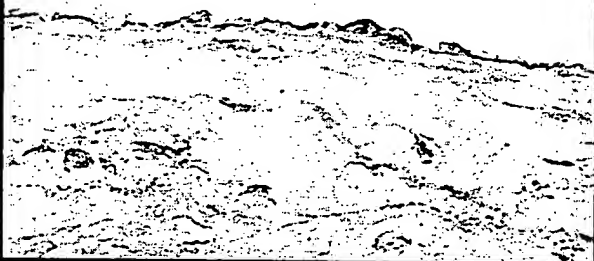


Fig. 4C



Day=3



Day=28

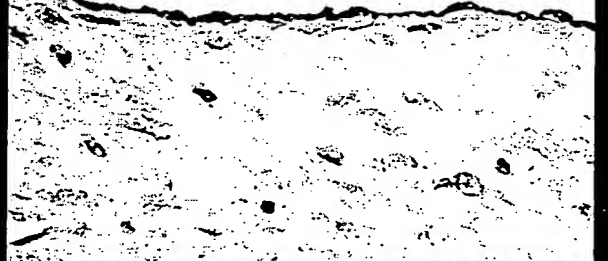


Fig. 4B

Fig. 4D

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T02250-EDBE9860

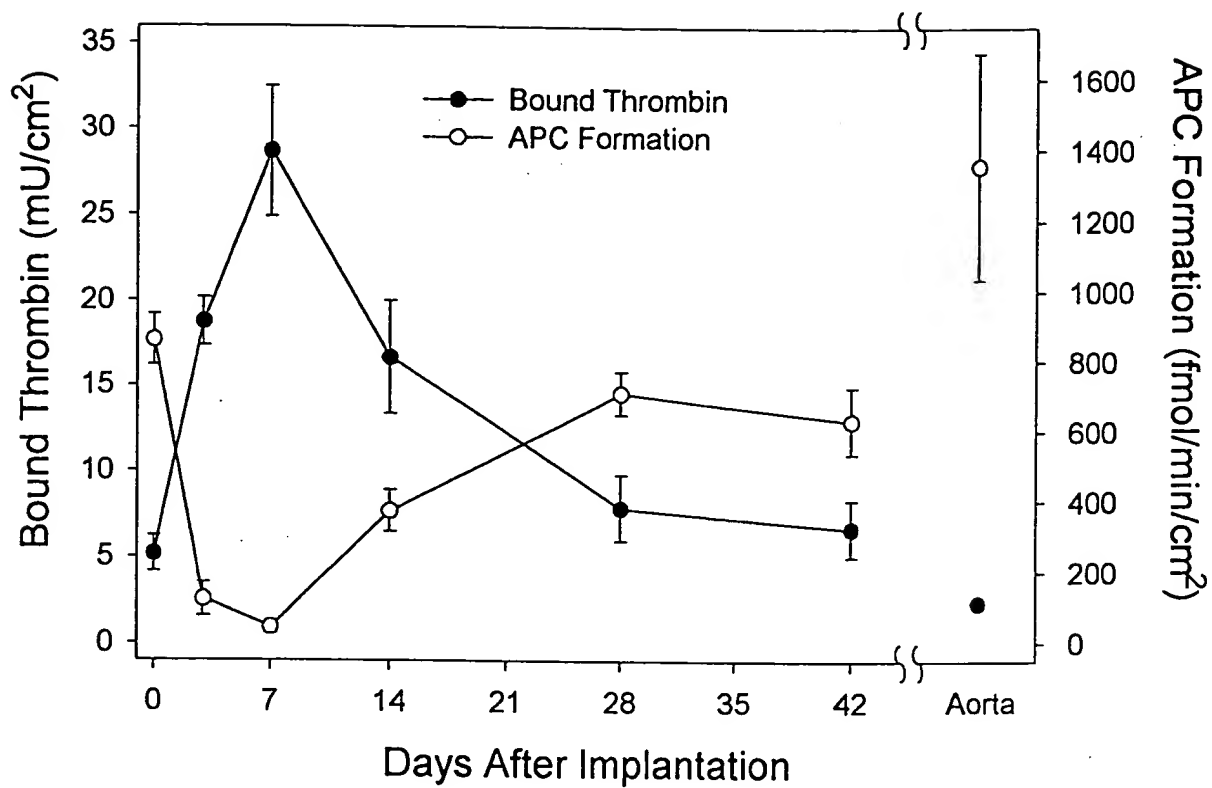


Figure 5

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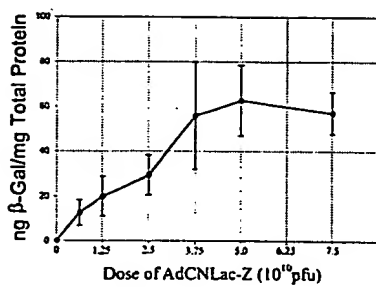


Fig. 6A

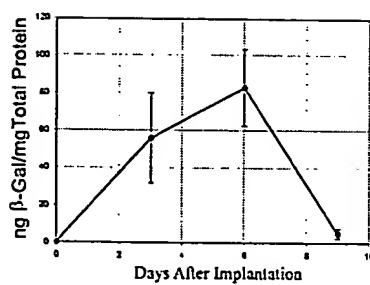


Fig. 6B

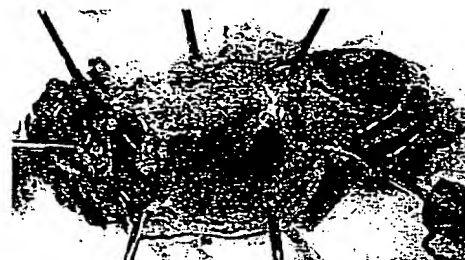


Fig. 6C

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The diagram illustrates the construction of a conditional gene expression system using Cre-loxP recombination. It shows two plasmids: a donor plasmid (top) and a pAdlox vector (bottom).

**Donor Plasmid (Top):** Labeled  $\Psi 5$ , it contains an ITR (Inverted Terminal Repeat) on the left, a loxP site (represented by a box with diagonal lines), a CMV Promoter, a loxP site (represented by a box with diagonal lines), and a gene labeled  $A_n$ . Below this, a linear DNA segment shows restriction sites:  $\Delta$  EI, BsaBI, BsaBI, BsaBI, and  $\Delta$  E3. A ClaI site is also indicated.

**pAdlox Vector (Bottom):** It contains an ITR on the left, a loxP site (represented by a box with diagonal lines), a CMV Promoter, a Multiple Cloning Site (MCS), a loxP site (represented by a box with diagonal lines), and another ITR on the right. The gene  $A_n$  from the donor plasmid is shown being inserted into the MCS of the pAdlox vector via recombination.

**Handwritten Note:** An arrow points to the MCS region with the text: "TM OR  $\beta$ -gal gene INSERTED HERE".

The diagram illustrates the Cre-mediated recombination strategy for generating a new virus. It shows the following components and steps:

- Donor Construct:** A linear DNA fragment containing an ITR (Inverted Terminal Repeat), a shaded region labeled "Exp. Cassette", and a loxP site, followed by a break in the DNA.
- Vector:** A circular plasmid labeled  $\Psi$  and  $\Psi 5$ , with a  $-polyA$  tag.
- Step 1:** CRE recombinase acts on the donor construct and the circular vector, resulting in a recombined intermediate construct.
- Intermediate Construct:** A linear DNA fragment containing an ITR, the "Exp. Cassette", and a pAdlox site, followed by a break in the DNA.
- Step 2:** CRE recombinase acts on the intermediate construct and another circular vector (labeled  $\Psi$  and  $\Psi 5$ ), resulting in the final "New virus" construct.
- Final Products:** The "New virus" construct, which contains the ITR, the "Exp. Cassette", and the  $\Psi$  and  $\Psi 5$  genome, and a separate ITR-loxP-ITR fragment.

Figure 7B

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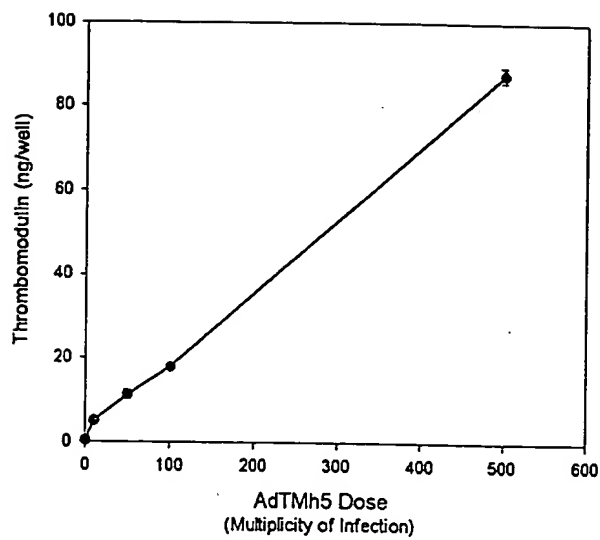


Figure 8A

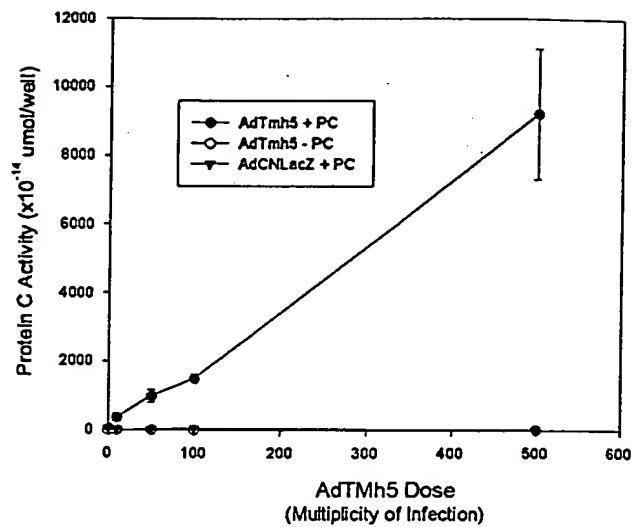


Figure 8B



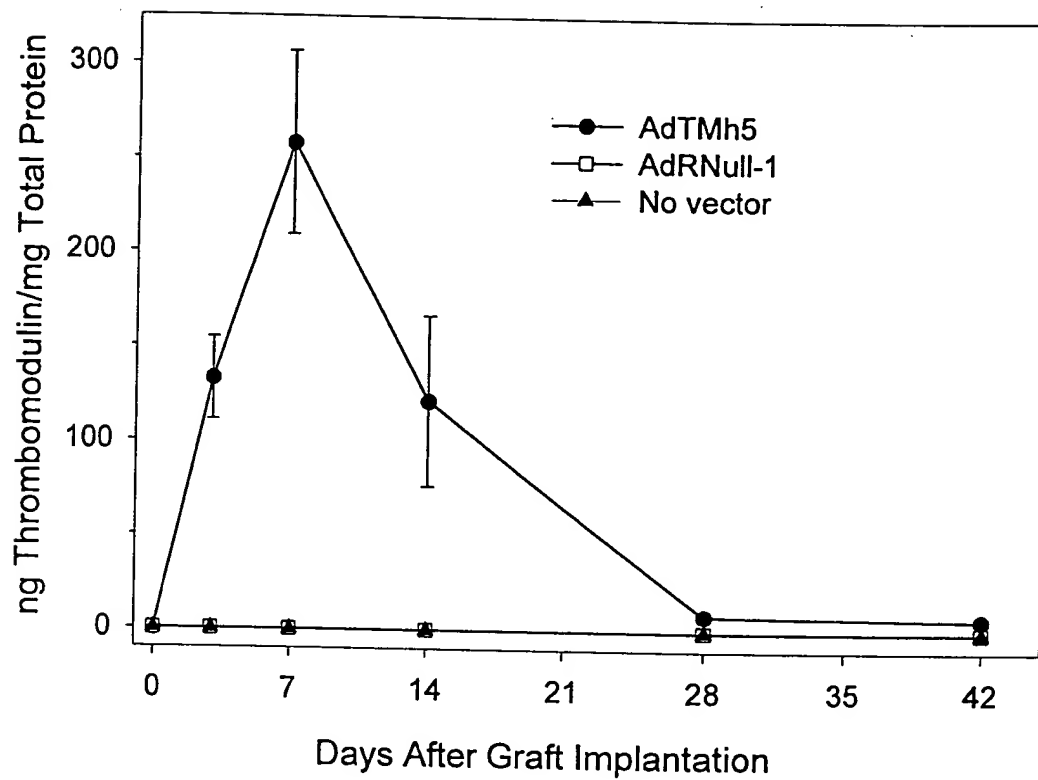


Figure 9

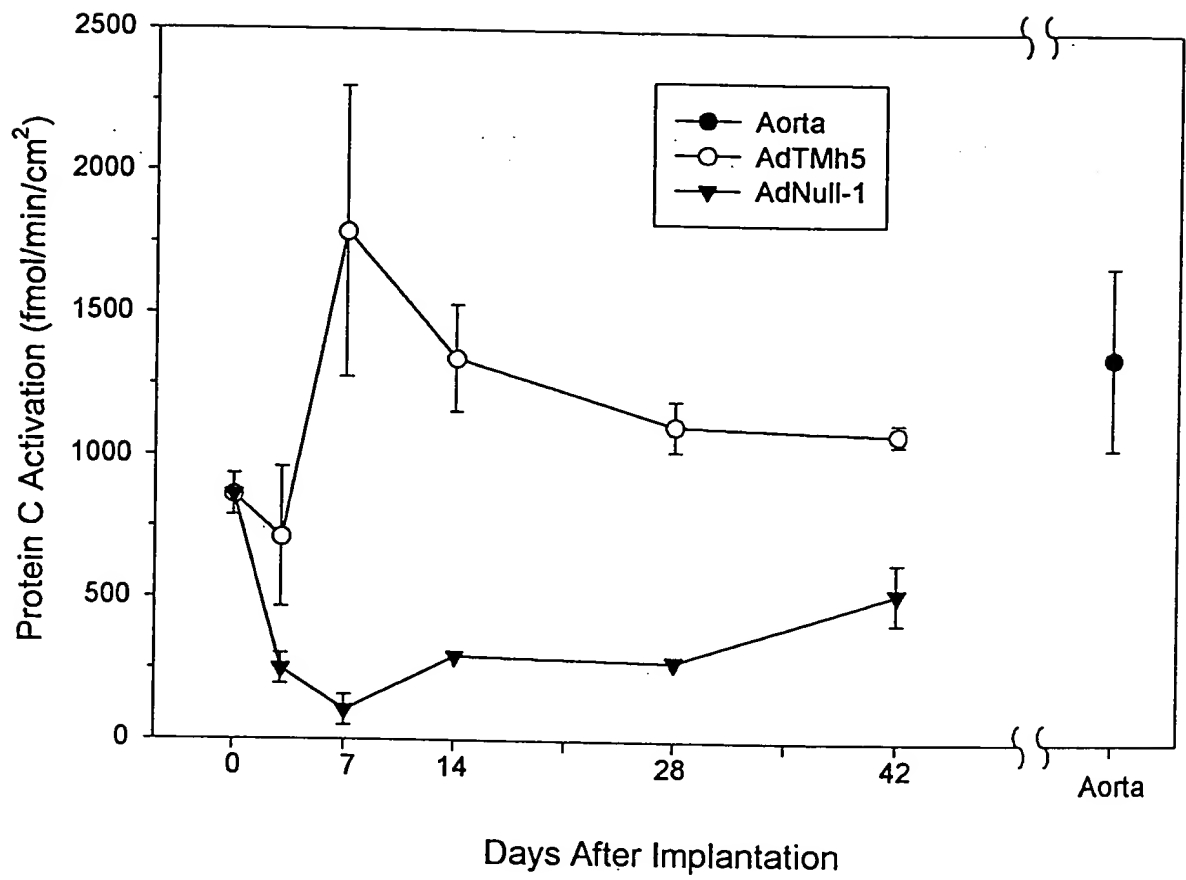


Figure 1.0

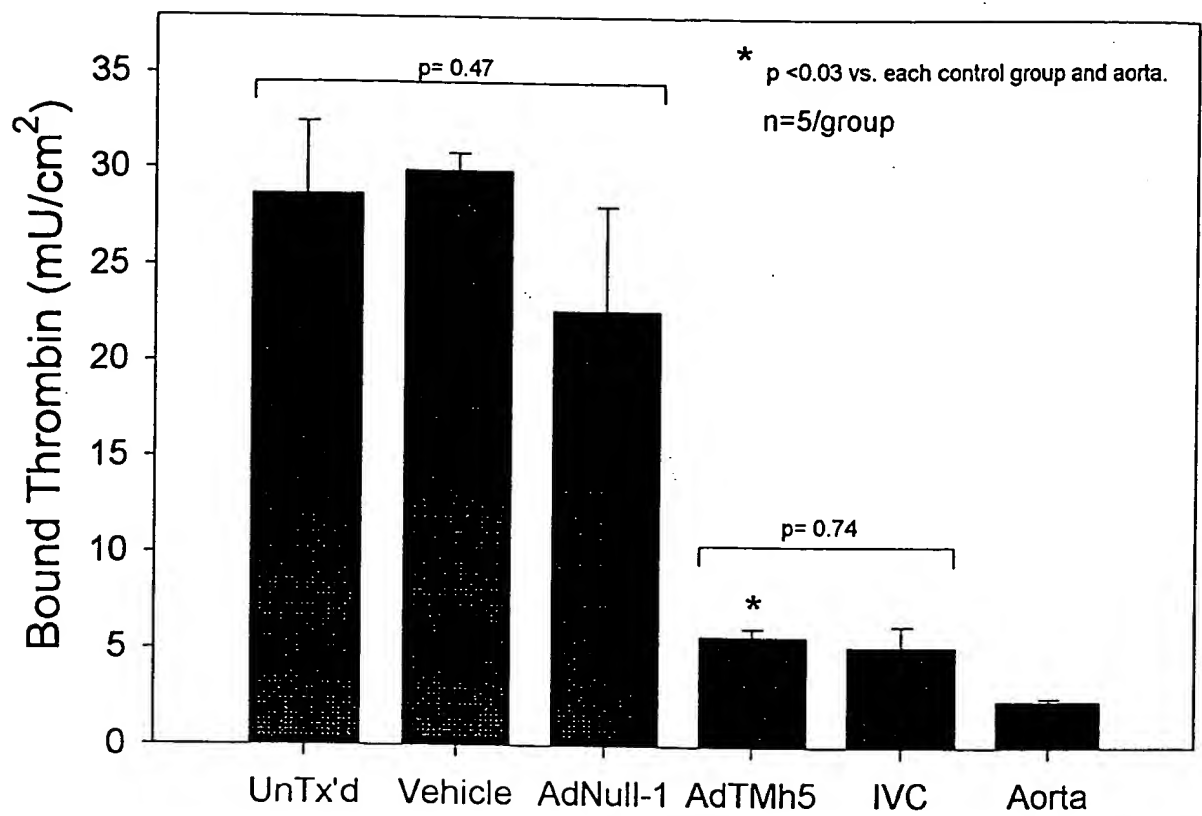


Figure 11

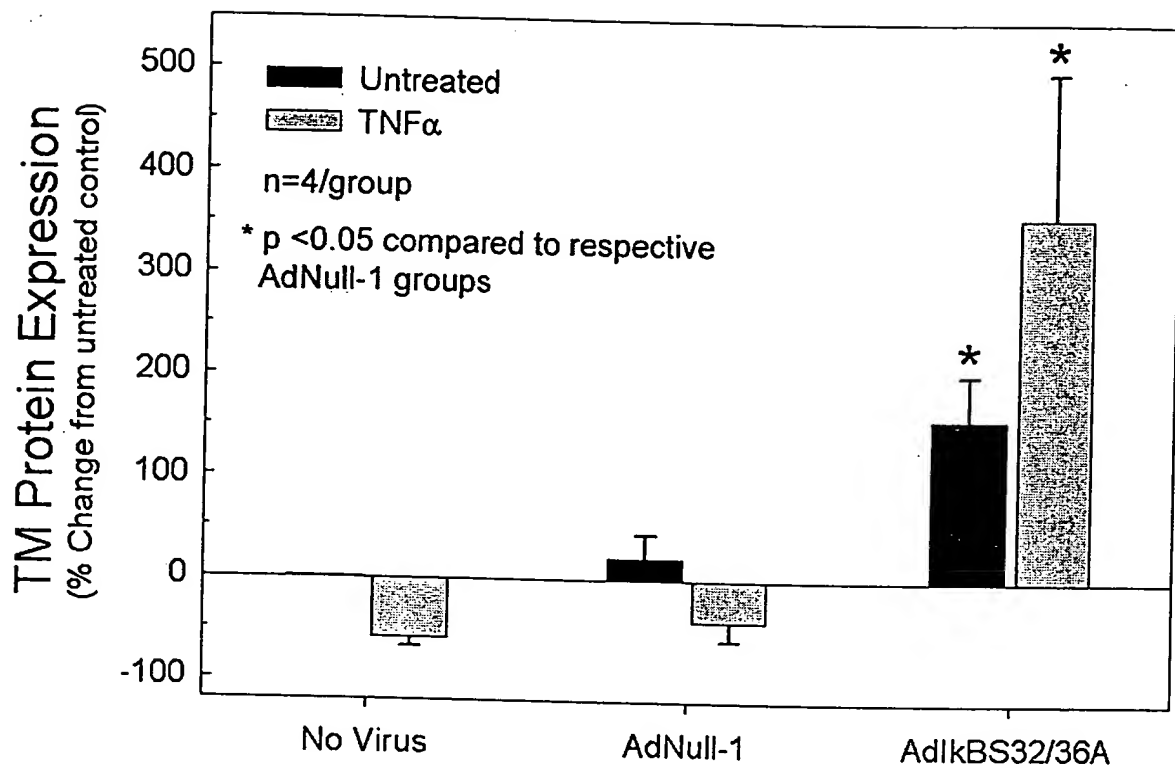


Figure 12